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THE PARASITE OF ORAL ENDAMEBIASIS, *ENDAMEBA GINGIVALIS* (GROS) *

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The pathogenic importance of the endamebae of the human mouth, which formerly were held as innocuous and were but scantily considered by pathologists, has been urged by the writers and others in a series of articles published within the past five or six months.[1] According to this view the writers regard these parasites as either directly causative of a large class of gingival and alveolar pyorrheas or as important members of a symbiotic chain with one or other of the numerous associated vegetable micro-organisms, in the production of these lesions. They find these endamebae also in the tonsils, in cases of chronic cryptal inflammation, often with tonsillar enlargement; and are disposed to regard them as concerned in the production or maintenance of such inflammatory changes of these organs; and moreover, believe that a long series of systemic complications of Riggs' disease and tonsillitis are due in some part to these parasites. Proof of the pathogenic importance of the endamebae in Riggs' disease rests upon their almost constant presence in the suppurating pockets of pyorrhea, and the prompt removal of both suppuration and of the endamebae when emetin, a proved amebicide but of low bactericidal value, is administered locally or generally. The authors hold that an overwhelmingly large group of cases of pyorrhea in which these endamebae are met should be segregated under the name "endamebic pyorrhea," in the same way that dysenteries in which dysenteric endamebae are met are known as "endamebic dysentery"; but reserve a small series of pyorrhea cases which cannot be included in this class. In attributing influence of these protozoa in the production of systemic complications the writers follow the general lines of argument as to the influence of "oral sepsis" of Hunter and others, merely emphasizing the endamebae as essentially involved because in their experience in a number of cases such complicating conditions have yielded, along with the oral lesions, to the use of emetin. Among such complications they include various chronic and recurrent arthritides (of the type of arthritis deformans), certain obscure anemias (as some of the splenic anemias and picked cases of pernicious anemia), functional and catarrhal disturbances of the gastro-intestinal canal, and probably, too, cases of degeneration of parenchymatous organs (as kidney and liver), an undefined group of serous inflammations (as of the heart), and perhaps, also, certain chronic functional nervous disturbances (neuralgias, etc.).

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* From the Laboratories of Pathology of the Medical School of the University of Pennsylvania, Philadelphia, Pa.

* Presented at Christmas Meeting of American Society of Bacteriologists, 1914.

1. Preliminary report by Barrett: Dental Cosmos, August, 1914. See also Smith: Dental Cosmos, September, 1914. Bass and Johns: New Orleans Med. and Surg. Jour., November, 1914. Smith, Middleton and Barrett: Jour. Amer. Med. Assn., November 14, 1914. Barrett: Dental Cosmos, December, 1914. Cf. Chiavaro: Abst., Dental Cosmos, September, 1914.

If no more of this comprehensive list of disturbances prove ultimately to be caused by these parasites than the local peridental suppurations and, perhaps, chronic cryptal tonsillitis, a careful systematic consideration of the parasites in question would be amply justified; and it is from this standpoint that the present discussion is presented, in an attempt to coordinate the amebiform organisms described by various observers as parasites of the human mouth and to compare them with other and better known forms of endamebae.[2]

In 1849 Gros[3] in a comprehensive article entitled "Fragments d'Helminthologie et de Physiologie microscopique," announced his discovery of an amebic parasite in the human mouth and published therewith drawings of the organism. There is little of importance, other than the announcement of the observation, in his text; but as the drawings are of importance, both are here reproduced (Fig. 1).

AMOEBEA GENGIVALIS

Au milieu des productions du tartre des dents, on voit des vibrios, une sorte de végétation qui est quelquefois très régulière; mais on n'avait pas encore mentionné les vésicules que nous avons représentées, Pl. VI, C. Ces vésicules ont un mouvement si lent et si obscur qu'il faut en étre averti pour remarque qu'elles prennent toutes les formes, par une extension et contraction amoebéenne, laissent toujours voir à l'intérieur des globules qui semblent se déplacer un peu, et étre l'analogue de ce que nous connaissons chez de certains infusoires soidisant polygastriques. Leur origine, leur rôle et leur fini sont ignorés. Elles se trouvent surtout à la face interne des dents. Est-ce encore une génération spontanée?

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[2] This spelling is adopted by the writers instead of *entameba,* proposed by Casigrandi and Barbagallo in 1897 (*Ann. d'Igenie,* vol. 7, p. 103), because in 1879, Joseph Leidy (*Proc. Acad. Nat. Sci.,* Philadelphia, vol. 31, p. 204), then professor of anatomy in this school, proposed the generic name *endameba* for a parasitic ameba of the cockroach (*endameba blattae*). Aside from a feeling of personal loyalty to Dr. Leidy the writers are impelled by the rules of priority to adopt his nomenclature.

[3] Gros: Bull. Soc. imp. de nat. de Moscow, Vol. 22, No. 2, pp. 549-573.

No one can look upon the drawings of Gros and fail to be convinced that he actually found amebae; and, if the size of the globules contained within them be accepted as that of the globular remnants of undigested leukocytic nuclei such as are commonly found in mouth endamebae, that the organisms were of about 0.025 to 0.030 mm. in diameter; that they threw out broad, rounded lobose pseudopodia and that in these pseudopods and at their bases a clear hyaline ectosarc was distinguished from a granular endosarc containing the coarse globular material. It may be accepted that these points, together with the habitat of the organisms upon the dental surface and in soft tartars were established by Gros.

In 1862, Steinberg,[4] in his thesis before the University of Kiev, in which he gives the details of his observations upon the living forms he encountered in the *substantia mollis alba* (soft tartar and refuse) about the teeth, probably refers, among other animal and vegetable organisms, to the same ameba announced by Gros thirteen years previously; but it is doubtful whether he had knowledge of the publication by Gros. At least he makes no mention of it, and writes as if under the belief that he is discussing a personally discovered amebic parasite, to which he attaches the name *amiba buccalis.*

The text is very curious but disappointing and there are no accompanying illustrations. However, because the volume containing his observations is rare in medical libraries and because of the difficulty of dealing with the original Russian language, the writers present the following translation, rendered for them by Dr. Fischelis of this city, of that part of the comprehensive article by Steinberg which concerns the matter in question:

AMIBA BUCCALIS

These organisms I met in various forms and sizes. Their characteristics are of a negative rather than of a positive character, in that they have no fixed size (this changing several times a minute); that they have no definite motile organelles (these being substituted by expansions or pseudopodia, appearing now here, now there, on different parts of the body); that they do not present a distinct direction of movement, as the pseudopodia by which they move appear at varying positions on the body and then retract again. The number of pseudopods is not fixed, now none, sometimes one, two, three, four and more; a change in the number of extensions occurring in the individual in a comparatively short time (one or two minutes). The shape of the pseudopodia in a given individual is not fixed, and their length is also variable. Granules are distributed within the body of the ameba and these do not retain any constant mutual relation, or to the individual itself or to the periphery of the ameba. I observed, however, a difference in the amebae seen on different days. While the size of the amebae in a given instance is constantly undergoing change, such change has its limitations; but it can be said that to-day, for example, the amebae observed are generally larger or smaller than those seen upon a previous day. Although the pseudopodia of a given ameba are in constant change, they may in one instance be more obtuse, short and rounded off, and in another larger, less obtuse and not rounded, etc. The body of the ameba with all its variability, depending upon the instability of the pseudopods, preserves a more or less common character or peculiarity, hard to describe; but the experienced eye of the observer notices and recognizes this peculiarity. In a given ameba, for example, there are always pseudopods present; in another there are times when none are to be seen. In a word, the total summation of a series of characteristics which are individually of no importance enables the observer to conclude, for instance, that he has before him not one single type of amebae but various amebae.

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[4] Steinberg: Souremenaya meditsina, Kiev, 1862, Nos. 21-24.

Having observed amebae in the soft white substance about the teeth and those in various infusions, I was able to convince myself that with all their indefiniteness of outlines and size there are certain features characterizing all amebae, viz., all change their positions by means of their pseudopods. Following the movements of various amebae I have always noted that first on some side of the body of the organism there appears a visible thickening, at first showing as a light and almost transparent cloud; this thickening (a pseudopod) grows more and more, eventually taking on a color nearly like that of the body of the ameba and then after one-half, five, ten, twenty minutes, when it has attained a certain size and form, it remains for a time fixed, as if it were adhering to the glass and becoming hardened in that position. Then the whole body of the ameba moves toward the point where the pseudopod has become fastened and coincidently the pseudopod diminishes, even to disappearance (at least it becomes invisible to the eye of the observer). During the time of the formation of the pseudopod, particularly toward the end of that period, the granules of various size in the body of the organism move toward the side where the pseudopod was formed. Sometimes, however, pseudopods are formed simultaneously in two or three places. Then they all begin to get smaller and the body of the ameba remains in an unchanged position. Or there is but one pseudopod, and when it has attained a certain size it begins to become smaller without the body of the organism having moved. Several times I measured the rapidity with which an ameba changed position, but found that this commonly varies; but it can be said that in one minute the body travels approximately from 0.001 to 0.003 mm. The amebae which I observed in infusions traveled in the same time as a rule considerably greater distances, although their bodies are not larger than that of the ameba of my own mouth. Probably this depends on the lower consistence of the infusions, since as the fluids evaporate the amebae begin to move more slowly. All the amebae which I have encountered in the soft white substance invariably present pseudopods of rounded outline; acute forms I have never noted in these, although I have seen this last form several times in the amebae of infusions. In the amebae of the mouth as in all amebae there is a considerable amount of more or less granular material enclosed in the body. In the midst of these granules there are one or two larger than the rest (nucleus?).

It may be said from these observations (1) that I have found these amebae comparatively few times, only six times in eighty-three observations (about 70 per cent.); (2) they are more frequently found on the internal surface of the teeth than on the labial side; (3) the amebae have been found only in the lower jaw; (4) once I observed them even after very careful cleansing of the oral cavity.

The only points of positive statement in his verbose attempt to describe what he observed, are that Steinberg saw in soft tartar motile amebiform organisms, with a small number and rounded type of pseudopods, the body contrasting with the pseudopods by its granularity and containing coarser or globular bodies. One or other of the largest of the latter he suspected was a nucleus. If he was correct in this surmise his *amiba buccalis* differs in its nucleus from that characterizing the ordinary examples of these mouth endamebae; if, as is probably the case, these coarse globules were merely remnants of ingesta, the nucleus in the parasites with which he was concerned was at least difficult to distinguish in the living organism. In the absence of direct statement it is worthless to speculate as to the size of his ameba. It is clear that the ectoplasm was hyaline and of a different "color" (refraction?) from the endoplasm, but there is no indication as to its proportionate amount save that it formed the pseudopods. It is of course utterly impossible to be positive because of the incompleteness of the description and the uncertainty of data; but at least the writers hold that these points are not incompatible with a belief that Steinberg's *amiba buccalis* was the same species of endameba as that now known and described more fully first by Prowazek under the same specific name.

In 1879 Grassi,[5] in a paper on parasitic protozoa, especially those of man, records (without illustrations) the finding of an ameba, which he named *amoeba dentalis,* in material obtained from gingivitis lesions. His brief text follows in the original:

> 4. *Amoeba dentalis* (mihi) (nell' uomo). E forse uguale alla *buccalis* di Steinberg (la memoria di quell' autore e rarissima e non potè consultarla neppure Leuckart). Ha molta somiglianse coll' *amoeba coli;* come essa al disotto dei 25° C. ò pochissimo mobile e tiene per lo più una forma tondeggiante; a 38° —40° C. è vivacissima e protea. Ne trovai numerosi esemplari in tre casi di ulite.

Later, however, although Perroncito also stated that he had observed these amebae twice in the mouth, Grassi doubted the verity of his observations, suggesting that the supposed amebae were perhaps "salivary corpuscles."[6] With our present knowledge there can be no doubt that Grassi's observation of amebae in the human mouth was correct; and the points in his brief notice that are especially noteworthy are that the parasites were obtained from cases of gingival inflammation (probably pyorrhea) and that they much resembled *ameba coli.* (This last statement, it must be remembered, was made before the present separation of the old species *a. coli* into *Endameba coli,* and *Endameba histolytica* obtained.) It is very significant, too, that by Grassi's counsel, and more or less under his direction, the dentist Chiavaro in 1914 devoted a study to *Endameba buccalis* Prowazek,[7] in which he states that "probably *Entameba buccalis* described by Prowazek should be identified with *amoeba gingivalis* Gros (1849), with *Entameba buccalis,* Steinberg (1862), with *Amoeba dentalis* Grassi (1879) and with the ameba found by Flexner in 1892 in Baltimore in an abscess of the lower jaw." Under such circumstances there seems little reason for extended discussion, and the writers accept in full the identity of Grassi's organism with that of Prowazek.

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[5] Grassi: Gazzetta med. Ital.-Lomb., vol. 39 (8th series, vol. 1), p. 446; Nov. 8, 1879.

[6] Grassi: Cf. Railliet, Traité de Zool., méd. et agricole, Paris, 1895, p. 118.

[7] Chiavaro: v. sup. for abstract; original, Ricerche sull' Entamoeba buccalis, Lavoro eseguito nel R. Instituto di Anatomia Comparata della Università di Roma diretto dal Prof. B. Grassi, Roma, 1914.

In 1904 Prowazek[8] announced his discovery of a parasitic ameba of the human mouth, under the name of *Entamoeba buccalis,* without reference to the earlier work above detailed, and repeating for his declaredly new species a name previously appropriated by Steinberg for an organism of the same genus and of the same habitat. His article is easy of access and forms the basis for the texts of prevailing works upon animal parasitology, and is confirmed in the writings of other observers;[9] for which reasons there is no need of detailing here more than the important data included in his original article. He met these organisms in material from the cavities of decayed teeth, first in Rovigno and later in several individuals in Trieste. He describes them as varying from 0.006 to 0.032 mm. in diameter when at rest; provided with a distinct ectoplasmic border, hyaline and refractive; a granular endosarc full of food vacuoles and ingested globular masses; mononucleated (the nucleus commonly deep in the body, small, 0.0015 to 0.0045 mm. in diameter, round, vesicular, poor in chromatin, with small, deeply chromatic "binnenkörper," a thick nuclear membrane refractive and of greenish tint and often showing chromatin granules collected along its inner limit); the whole nucleus of firmer consistence than that of *Endameba histolytica* Schaudinn and not compressible by the coarse ingested masses in the endosarc; endowed with active motility; pseudopodia few, broadly lobose and rounded, but sometimes forming an elongation of the animal as a single broad pseudopod; without contractile vacuole. He speaks of the frequent presence of residua of leukocytic nuclei in the nutrition vacuoles; and describes reproduction by simple division after nuclear swelling, mitotic changes in the binnenkörper and division of the nucleus. He also calls attention to what he believes to be chromidia of nuclear origin in the cytoplasm, which he suggests may develop into complete nuclei of young amebae (which then separate from the parent by gemmation); but at time of publication he has not met evidence of reproduction encystment. He recognizes a general resemblance to *Endameba coli* (Lösch) and *Endameba histolytica* Schaudinn, but differentiates it from the former by its clearer and complete hyaline ectosarc and by the differences in the observed modes of reproduction; and from the latter mainly by the greater rigidity of its nucleus and more definite structure of the nuclear membrane.

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[8] Prowazek: Arbeiten aus d. Kais. Gesundheitsamte, vol. 21, p. 42.

[9] Cf. Leyden and Löwenthal: Charite Annalen, vol. 29, p. 3, in a case of cancer of mouth.

These characters in a general way the writers can corroborate, but in some points would insist upon a greater latitude of variation than the outline given above would indicate. And in fact Prowazek in his article, in commenting upon the completeness of the ectosarc, specifically indicates much variation in its thickness; and, in speaking of the rigidity of the nucleus, acknowledges that in rare instances when it is, as occasionally may be noted, in eccentric position, close to the ectosarc, it may show indentation in the varying movements of the animal. Leyden and Löwenthal[9] who identify their organisms with that described by Prowazek vary from his statements as to the size (0.008 to 0.020 mm.) of the organism, the uniformity of distinction of ectosarc and endosarc and as to the position of the nucleus (sometimes at border of endosarc and ectosarc but usually central); and they fail to recognize in the living animal the peculiar greenish shimmer mentioned by Prowazek in the nuclear membrane, suggesting the operation of the personal equation in the observation. In our earlier papers upon this subject the writers have held that of the endamebae of the various cases of pyorrhea examined all but one or two (tentatively held as *Endameba kartulisi* Döflein) were examples of *Endameba buccalis* Prowazek, but suggested that perhaps fuller study might indicate that of the larger group some should be referred to a third, unidentified species. This last point was based upon the fact that occasional examples varied beyond the usual size, attaining as much as 0.038 or 0.04 mm diameter, that these were apt to show a greater general activity of movement, and often extruded unusual numbers of pseudopods and these of unusual shapes (often small drop-like projections, or small projections of the same type upon the border of the main pseudopods). Further study has led us to believe these features are not of specific differentiating value, and as the other characters, studied in the stained specimens, are identical we are of the opinion that such forms are merely exceptional individuals of one general species. While in a given material some examples may have attained the size mentioned, the greater number were smaller; and while in a given observation a greater activity of the amoebae of a certain individual was noted, a second preparation on the same or a subsequent day from the same person has repeatedly failed to show the same degree of motility. And in one and the same ameba we have repeatedly seen alternate periods in the course of a brief observation of the living animal when the pseudopods would vary from one or two large, rounded lobose projections, to longer and digitate types, to be succeeded perhaps in a short while by numerous small drop-like projections of the ectosarc. For such reasons we are satisfied occasional variations, even if marked, cannot be insisted upon for specific identification, and that only the common type, as the mean number, size and form of the pseudopods, should be held as of value in this connection. With this preface our description of these parasites may be outlined as follows: Naked parasitic amebae of usual diameter in resting examples of 0.030 to 0.035 mm. (with exceptional instances reaching 0.040 or slightly above); with refractile and faintly greenish-tinted hyaline ectosarc well defined from the granular endosarc, but sometimes so thin as to be easily overlooked; endosarc granular, colorless and in all but the more minute examples containing few to many digestion-vacuoles in which globular detritus of leukocytic nuclei and red blood cells are commonly found along with bacteria; with a small (0.002 to 0.005 mm.) rounded nucleus, invisible or at best uncertainly distinguishable in the unstained specimen, usually central or subcentral in position (Fig. 3; a, b), but at times eccentric (when the ingesta push it to one side) (Fig. 3; c, d, e). The difficulty of distinguishing the nucleus in the living animal prevents the writers from expressing a positive opinion as to its greater rigidity than that possessed by the nucleus of *Endameba histolytica* Schaudinn, but we have certainly met in stained specimens oval and slightly indented nuclei which makes us hesitate to accept this point which Prowazek especially indicates as distinctive from the dysentery endameba (Fig. 3; f, g, h). The nucleus is very poor in chromatic substance, vesicular, with a small "binnenkörper," sometimes showing a minute centriole, a clear space between it and the nuclear border containing no chromatin or at most a very few incomplete threads; and the border is represented by a thin but somewhat irregular line of chromatin, about which the writers are unable to recognize a further membrane and which they regard as the membrane itself. The thickness of this membrane is not absolutely fixed, sometimes more delicate, sometimes less so, but always thin, with scattered parts thicker than others or with scattered chromatin granules on its inner surface (Fig. 4). The degree of motility manifested by the parasites is to us fairly comparable to that of the dysenteric endameba (Fig. 5). The pseudopods as a rule are few (one, two, or three), usually broadly lobose and commonly attaining a maximum length of the diameter of the endosarc. Not infrequently a single extension may reach even a greater length, making the full long measurement of the parasite 70 or more micromillimeters. Or occasionally there are more pseudopods, these then small and guttulate. The pseudopods are composed practically entirely of the ectosarc, the granular endosarc terminating at the base, or extending but a little into the larger ones. We have observed examples containing two nuclei (Fig. 3; i) and have seen binary division of the living ameba take place; and have repeatedly seen small protoplasmic masses separate by gemmation (some of these containing minute particles of chromatin); although we are uncertain as to definite chromidial formation, as we are confused by bacteria and bacterial fragments which are numerous in the larger amebae and which stain very like the chromatin with Giemsa stain and with iron-hematoxylin which we have usually used for study. We have found "dauer" cysts, but thus far no reproduction cysts.

With these data in hand the writers do not hesitate, in spite of the published differences (which we believe are due to nonspecific variations), to identify the organism with which we have become fairly familiar, with that of Prowazek. As above indicated, we see no reason to believe the *Ameba dentalis* of Grassi to be other than the same species, if we may accept identity of habitat, identity of lesion, comparable morphology (as expressed in terms of the dysenteric endameba), and the evident later opinion of Professor Grassi as to identity (inferable from his relation with Chiavaro's paper). It is impossible to be sure of Steinberg's *Amiba buccalis;* but identity cannot be denied or specifically asserted. The *Amoebea gengivalis* of Gros is so nearly like the same organism, as shown by his drawings and a few points in the text, that it is impossible to say it is not the same and quite within reason to say that it probably is identical. Gros may therefore be held as probably the original observer of the common oral endameba with which we are today familiar; and by rule of priority, with modification of spelling of *gengivalis* to *gingivalis* to conform with etymological propriety this organism should take the nomenclature *Endameba gingivalis* (Gros) instead of the more common present-day name *Entameba buccalis* Prowazek. In this attitude in fact the writers are not the first.[10]

In addition, the writers feel that further attention should be given to certain amebae which have been met by Kartulis,[11] Flexner,[12] Verdun and Bruyant[13] and Bruyant and Pelissier[14] in the same comparative lines. The first and second refer to amebae met in the pus of abscesses of the lower jaw, the third to organisms in the pus of two symmetrical abscesses of the cheeks, and the fourth to amebae in pus from suppurative gingivitis. Flexner's ameba was regarded by himself as indistinguishable from the dysenteric ameba (*endameba histolytica?*), an organism apparently with no marked disdistinctness of differentiation between the homogeneous ectosarc and the granular endosarc, the latter containing numerous vacuoles and showing in its interior red blood cells and their detritus. The nucleus was not recognized in the living cell; the pseudopods were characteristically blunt, varying from a mere bulging to a change in which the greater part of the ameba was protruded. No actual measurements were published by the observer, and no drawings accompany the text. It is usually classed as *endameba Kartulisi.* General conformity to the characters above indicated for *Endameba gingivalis* (Gros) seems to the writers, however, to be included in these incomplete data. That *Endameba gingivalis* (Gros) may be met in such maxillary abscesses the writers can testify positively on the basis of two cases. One of these occurred secondarily to alveolar fracture occurring in the extraction of a carious tooth, and was accompanied in its course of three or four years by the discharge of sequestra from the jaw and extension of the suppuration to the surrounding soft parts in the cheek and floor of the mouth, with establishment of an external fistula. At the time of discovery of the amebae in this case the fistula had healed, but sequestra were from time to time being discharged through the gingival tissues into the mouth. The mouth was kept scrupulously clean by the patient and no amebae were detected about the remaining teeth or over the exposed surface of a small sequestrum which was gradually being dislodged into the mouth; but when this was extracted numerous amebae of the ordinary *gingivalis* type were detected in spreads from its embedded surfaces. The case was treated with emetin and has promptly and completely cleared up. The second case was regarded as a dental cyst of the lower jaw, but found by the surgeon to be a chronic maxillary abscess in the pus of which the writers found large numbers of endamebae which were as a rule of larger size than usual, quite active, very commonly contained red blood cells, but on close study of stained specimens were also identified with the ameba of Gros. There seems but little reason, therefore, to regard the organisms of Flexner's case as specifically different. The impression has grown upon the writers that the ameba of Flexner became associated with that of Kartulis into an isolated species mainly because these cases of Flexner and of Kartulis were both of suppuration of the lower jaw, both published at nearly the same time and appeared before any detailed attention had been given to oral amebae, Prowazek's study not being brought forward until a decade later. Prowazek, at the close of his article above referred to, promised a further and fuller description of his organism with differentiation from the organism of Kartulis; but the writers have failed to find this later article if it be in existence.

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[10] Brumpt: Précis de Parasitologie, 1st and 2d editions, Paris, 1910 and 1913.

[11] Kartulis: Ztschr. f. Hygiene, 1893, vol. 13, p. 9; and Kolle u. Wassermann: Handb. d. pathogen. Mikroorg., 1st ed., 1907, vol. 1, p. 356.

[12] Flexner: Johns Hopkins Hosp. Bull., November, 1892, vol. 3, p. 104.

[13] Verdun and Bruyant: L'Echo méd du Nord., Aug. 11, 1907, vol. 11, p. 375.

[14] Bruyant and Pelissier: L'Echo méd. du Nord., June 29, 1909, vol. 13, No. 26.

In 1893 Kartulis[15] announced his discovery of amebae in the pus from an abscess of the lower jaw (Fig. 6), and subsequently[16] reported similar findings in five more cases of suppurative osteomyelitis of the jaw. These organisms were named in honor of the discoverer by Döflein *Endameba kartulisi,* prior to the publication of the specific name *maxilaris,* given by Kartulis (the latter now appearing as a synonym). In all three references (*supra*) to these observations of Kartulis his first impression of similarity to the dysenteric ameba is noted. Differentiation is made because of the somewhat larger size of the organism and its higher degree of activity, both progressive and pseudopodial. In the original description, too, the nucleus is declared to be smaller and thus not comparable to the nucleus of the organism of dysentery; but it must be recalled that at the time *Endameba coli* (Lösch) was still included with the species now known as *Endameba histolytica* Schaudinn, and comparison with the larger nucleus of the former (readily observed in the unstained specimen and staining more deeply) might fairly be the cause of this statement. Kartulis describes the nucleus as small, rarely distinguishable in the unstained specimen, vesicular, and provided with an easily seen nucleolus (in stained state). It is said to be surrounded by a clear zone, referring apparently to a finely reticular perinuclear area of the endosarc. Comparing the text cuts and the colored plate illustrations of the original article by Kartulis, the diameter of the nucleus (not definitely stated) must be about 5 or 6 micromillimeters; and while in both the text cut and the colored plate it would appear to be a relatively solidly chromatic body, the "vesicular" character described indicates that the "nucleolus" is a small "binnenkörper" surrounded by a relatively clear plasm, about which a probably fairly marked nuclear wall occurs separating the intranuclear substance from the light reticular perinuclear part of the endosarc. The ectosarc is described as clearly seen in the pseudopodia, but (in the resting stage) not apparent as a peripheral zone. Both in the original text and in the cuts the pseudopods are indicated as few, large and digitate (in the later descriptions said to vary from *lobose shape* to long and comparable to the "horns" of a snail), reaching perhaps several times the length of the body diameter. If the above interpretation of the nuclear picture be correct the points of distinction from *Endameba gingivalis* (Gros) rest more particularly upon the

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[15] Kartulis: Ztschr. f. Hyg. u. Infectionskr., vol. 13, p. 9.

[16] Kartulis: Centralbl. f. Bakt. u. Parasitenk. 1 Abt., Ref., vol. 33, p. 471; 1 Panhellen. Med. Congress in Athens, May 6-11, 1901; see also Kolle u. Wassermann, Handb. d. pathogen. Mikroorg., 1st ed., vol. 1, 1907, p. 356.

EXPLANATION OF PLATE

Fig. 1.—Photographic reproduction of that portion of the plate of illustrations accompanying article by Gros in 1849, which depicts his *Amoebea gengivalis.*

Fig. 2.—Photographic reproduction of part of plate illustrating article by v. Leyden and Löwenthal upon *Endameba buccalis* Prowazek, showing organisms in motion.

Fig. 3.—Camera lucida drawings of *Endameba gingivalis* (Gros), stained with iron hematoxylin; *a* and *b* showing the usual central or subcentral position of the nucleus; *c, d* and *e,* examples with the nucleus in eccentric position; *f, g* and *h,* examples showing nucleus in compressed condition; *i,* an example with two nuclei (it is suspected, although not known, that the small ameba lying within the same space in the stained film had been recently separated from the larger one.)

Fig. 4.—Photomicrograph of *Endameba gingivalis* (Gros), stained with iron hematoxylin; from material from pyorrhea pocket.

Fig. 5.—Composite outlines of moving *Endameba gingivalis* (Gros), including five camera lucida sketches; time included, twenty seconds; to show activity of movement and long type of pseudopod at times assumed; magnification as in Figure 3.

Fig. 6.—Photographic reproduction of a text cut of *Endameba kartulisi* Döflein from original article by Kartulis in 1894.

Fig. 7.—Photographic reproduction of text cut illustrating article in 1907 by Verdun and Bruyant, showing *Endameba pyogenes* Verdun and Bruyant.

Fig. 8.—Photographic reproduction of cut of endameba described by Ribbert, here shown in parotid duct.

Fig. 9.—*Endameba mortinatalium* Smith and Weidman, in minute hepatic abscess; stained with iron-hematoxylin.

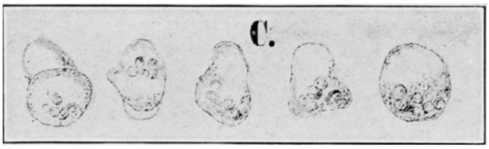

Fig. 1

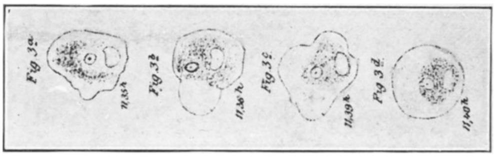

Fig. 2

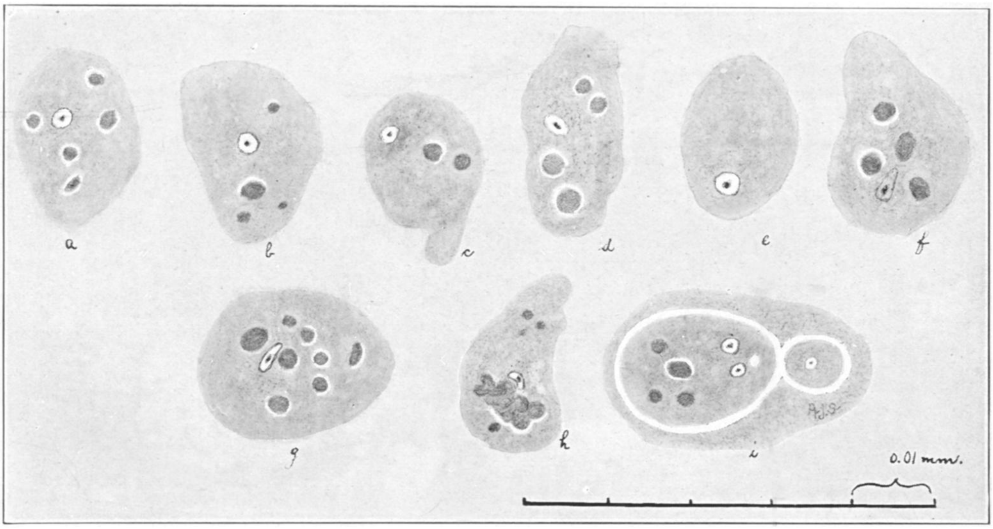

Fig. 3

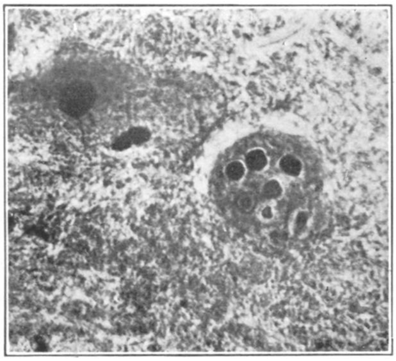

Fig. 4

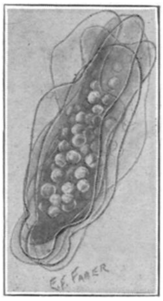

Fig. 5

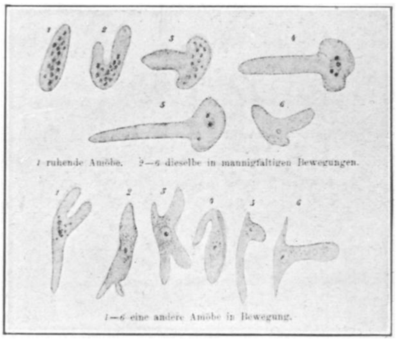
1
2
3
4
5
6
1 ruhende Amöbe. 2—6 dieselbe in mannigfaltigen Bewegungen.
1
2
3
4
5
6
1—6 eine andere Amöbe in Bewegung.

Fig. 6

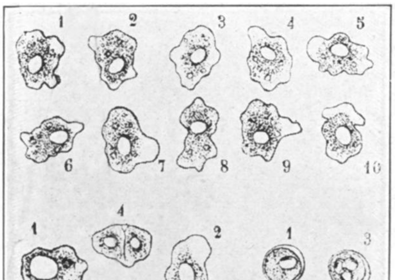

Fig. 7

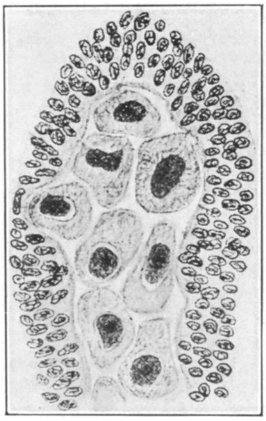

Fig. 8

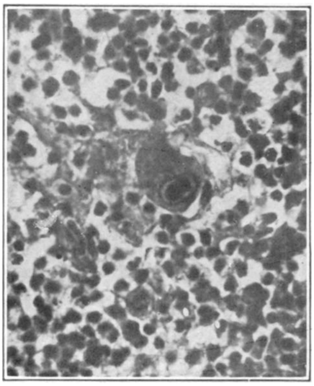

Fig. 9

size of the organism, the size and shape of the pseudopodia, the activity of movement and the lack of a clear ectosarcous border—in all of which particulars nonspecific variations of at least comparable degree may occur. Certainly in the occasional organisms which we have in the living state suspected of showing the characters of the ameba of Kartulis because of these latter points, the writers have seen the individual change and assume the general features of *Endameba gingivalis* (Gros), and subsequent study of stained examples has led us to conclude identity with the latter. We are of opinion, therefore, that there is not sufficient basis for accepting the specificity of Kartulis' type of these amebae, and would refer it also to the species *gingivalis* of Gros.

There remains of this group of parasitic amebae of the mouth for comparison the organism known as *Endameba pyogenes* of Verdun and Bruyant,[17] studied by these writers in the pus from two symmetrical abscesses of the cheeks in a case under the surgical care of Dubar and Leroy, in which there had preceded the abscesses carious teeth and pyorrhea lesions, to which the clinicians were disposed to attribute the abscess development (Fig. 7). Later Bruyant and Pelissier[18] mention the discovery of similar amebae in two cases of gingival suppuration, but with no more than partial description refer for their characters to the original article by Verdun and Bruyant. The mean diameter of these parasites is given as from 0.03 to 0.035 mm. They are described as quite active in movement, with large rounded pseudopodia rapidly projected and retracted. The pseudopods are hyaline in appearance, the endosarc granular; the ectosarcous border is not clearly seen, being absent or only visible in the pseudopods and their immediate bases. The endosarc is commonly full of digestive vacuoles in which remnants of erythrocytes and leukocytes are to be detected. The nucleus is described as relatively large, from 0.008 to 0.015 mm. (from one third to one half the diameter of the resting ameba), always visible, granular but paler than the surrounding endosarc. Double contoured cysts, from 0.006 to 0.015 mm., are described and said to frequently contain multiple, but never more than four, nuclei. Larger cysts, with single wall and a single unchanged nucleus are described, and perhaps are resistance encystments. Stained with Borrel blue and eosin, the body of the ameba is light blue, the nucleus (distinguished from the deeper blue remnants of leukocytic nuclei and copper-red residua of red blood cells about it) is apparently stained diffusely a reddish violet tint; is granular and often shows a well-defined nucleolus (binnenkörper). In brief, the size of the nucleus, its richness in chromatin and its visibility in unstained specimens, along with the mode of reproduction within a cyst, mark this as a species of these mouth amebae distinctly different from those hitherto discussed in this paper; and as such it must be kept clearly in mind in studies of oral endamebiasis.

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[17] Verdun and Bruyant: L'Echo méd. du Nord., Aug. 11, 1907, vol. 11, p. 375.

[18] Bruyant and Pelissier: L'Echo méd. du Nord., June 27, 1909, vol. 13, No. 26.

It is impossible to come to a definite conclusion as to the relationship of this last organism with certain other endamebae which have been described from the human body which contain large and richly chromatic nuclei because of the deficiency of data on both sides, but it may be suggested that in this connection there should be remembered certain organisms described by Smith and Weidman[19] under the name *Endameba mortinatalium,* being found by them in small abscesses in the liver and kidneys and also in the lungs of a still-born fetus (non-syphilitic). This these writers identify in a later note (October, 1910) with unnamed organisms met by Jesionek and Kiolemengolou[20] in the kidneys, liver and lungs of a syphilitic eight months fetus, and also by Ribbert[21] in the renal tubules of a syphilitic new-born infant twenty years before his publication, and twice thereafter in the parotids of nonsyphilitic infants (Fig. 8). Tietze[22] also records the discovery of amebiform organisms in a parotid tumor in a child of about 4 years of age; but these were held by Schaudinn, who examined histological sections, to be either identical to or akin to Prowazek's endameba. Smith and Weidman[23] have recently encountered the same large-nucleated ameba in the lungs of a congenitally syphilitic baby. These organisms (Fig. 9) in fixed and stained state, for the most part measured 0.025 to 0.032 mm. in diameter; the nucleus spherical or oval and usually from one third to one half the cell diameter. Considerable variation exists in the amount of ectosarc, which however in some examples is seen as a complete clear border zone in the stained examples. The endoplasm is highly granular, containing at times red blood cells and leukocytic detritus, and in some instances is full of chromidia-like bodies. The nucleus contains a very large, granular and richly chromatic binnerkörper, occasionally showing a centriole; about the binnenkörper a narrow clear zone of nuclearplasm and a delicate chromatin-staining nuclear wall, with threads or grains of chromatin scattered in the clear zone. Pseudopodia few and rounded in the stained specimens. The occurrence of these organisms in the parotid and their general comparability suggest that further study of relation be made.

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[19] Smith and Weidman: Univ. of Penna. Med. Bull., September, 1910.

[20] Jesionek and Kiolemengolou: München. med. Wchnschr., 1904, No. 43.

[21] Ribbert: Centralbl. f. allgem. Pathol., 1904, vol. 15, p. 945.

[22] Tietze: Mittheilung. aus d. Grenzgebiet. d. Med. u. Chir., 1905, vol. 15, p. 303.

[23] Smith and Weidman: Am. Jour. Trop. Med., vol. 2, p. 256, October, 1914.

Finally the writers (with natural hesitation because of full appreciation of the resultant confusion in our current views as to the relations of endamebae to dysentery and the impossibility of satisfactorily explaining a great field in the etiology and pathology of endamebic dysentery and its immediate complications as the liver abscess, if this statement be thought worthy of consideration) feel constrained to acknowledge that from a purely morphological standpoint we are unable to differentiate the organism which we believe to represent the vast majority of oral endamebae and to occur in an extremely large number of persons not merely in the tropics but all over the world, from *Endameba histolytica* Schaudinn. We are unwilling to make any assertion which involves biological identity in full, merely asserting that the morphological similarity is so close that we feel unable to make a distinction from microscopic observation alone. The limits of size of the two organisms are too close to permit this to serve as a specific difference; in both (Fig. 3; a, b, c, d, e) there is a tendency to clear ectosarcous differentiation from the granular endosarc in which cellular detritus and especially red blood cells and their debris may be seen; in both there is a small nucleus (the position of which is not fixed for either) of vesicular type, poor in chromatin, practically invisible in the unstained specimen, and showing the same small binnenkörper, the same plasmatic space between the latter and the nuclear border, which is formed by a delicate but variably irregular and chromatic nuclear membrane; both show few pseudopodia characteristically, and these usually of broad lobose type; and the general type of motility of both the cell and the pseudopods is closely comparable in the main but variable in both. Both forms, too, divide by binary fission and by gemmation,[24] both fail to form reproduction cysts as far as is known. And if it be a characteristic of *Endameba histolytica* Schaudinn to destroy red blood cells, so, too, is *Endameba gingivalis* (Gros) capable of rapid digestion of these same elements. We have repeatedly watched the complete disappearance of freshly englobed erythrocytes in the living ameba of Gros, the red cell fading before our eyes to invisibility in from three to five minutes.

Prowazek in his original article compares his ameba, which we have above held to be identical with *Endameba gingivalis* (Gros) with

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[24] *Note by Editor:* Gemmation, as a form of division in *E. hystolytica,* has been proved not to occur, the so-called gemmation being due to degenerative changes. In *E. histolytica* reproduction cysts occur, of course; the four-nucleated cyst of this species being easily distinguished from the eight-nucleated cyst of the harmless *E. coli.* If it be true that reproduction cysts do not occur in *E. gingivalis,* this alone would serve to distinguish this species from *E. histolytica.* Likewise the *tetragena* type of nucleus of *E. histolytica* does not occur, according to the author's description, in *E. gingivalis.*

Endameba histolytica Schaudinn, acknowledging its close conformity, but differentiating it by the point that its nucleus is more rigid, less likely to present compressed, flattened outlines. Such a point is exceedingly questionable; nuclear deformation may depend as much upon the amount and rigidity of ingested substances and the movement pressure of the cell as upon any special nuclear consistence. Prowazek from his text gives the impression that this is best appreciated in the living and moving organism; but it is difficult to observe the nucleus of either one in this state. Occasionally at least the writers have met slightly indented and oval nuclei in stained specimens of *Endameba gingivalis* (Gros).

Even if this suggestion be refused, the writers feel there is need of a more easily demonstrable differentiation, and believe that more than merely morphological studies are requisite to prove the dual specificity.